Genetic Resources and Crop Evolution **46:** 611–618, 1999.

Oil and meal characteristics of core and non-core safflower accessions from the USDA collection

R.C. Johnson[1], J.W. Bergman[2] & C.R. Flynn[2]
[1]*Western Regional Plant Introduction Station, USDA-ARS, 59 Johnson Hall, Washington State University, Box 646402, Pullman, WA 99164-6402, U.S.A. (E-mail: rcjohnson@wsu.edu);* [2]*Montana State University, 1501 North Central Avenue, Sidney, MT 59270, U.S.A. (E-mail: jbergman@sidney.ars.usda.gov)*

Received 17 December 1998; accepted in revised form 15 March 1999

Key words: Carthamus tinctorius, core collections, fatty acids, phenolic glucosides, safflower, vitamin E

Abstract

Germplasm evaluation of *ex situ* collections is needed to document collection characteristics, enhance utilization, and to determine collection needs. The objectives of this study were to (1) provide oil and meal evaluation information for a major portion of the United States Department of Agriculture (USDA) safflower (*Carthamus tinctorius* L.) collection, (2) compare ranges, variances and means between 203 core and 797 non-core accessions, and (3) determine if region of origin could be differentiated based on accession oil and meal characteristics. Means of the core and non-core accessions differed for % oil, palmitic acid, stearic acid, α-tocopherols, and phenolic glucosides (both bitter and cathartic) ($P<0.05$). Differences between linoleic acid, oleic acid and β-tocopherol means were not significant, and the variance between core and non-core accessions differed only for palmitic acid. Thus the core was not fully representative of the non-core accessions, but did capture a large fraction of the diversity in oil and meal factors present. Accessions from the Americas, China, South-West Asia, and South-Central Asia were differentiated using canonical discriminate analysis, but these regions overlapped to varying degrees with the E. European, Mediterranean, and E. African regions. Variation in % oil and fatty acids were generally more important than tocopherols and phenolic glucosides in differentiating accessions on a regional basis. The check cultivars Montola 2001 (high oleic) and Morlin (high linoleic) had oil and fatty acid content comparable to the maximums found in the collection. The ranges for tocopherols and phenolic glucosides among collection accessions, however, exceeded those of the cultivars, suggesting that collection accessions could be useful for genetic manipulation of these factors.

Introduction

Safflower (*Carthamus tinctorius* L.) is one of our oldest domesticated crops. Weiss (1971) reports that safflower probably originated as a crop in the Euphrates basin and was certainly grown more than four thousand years ago in Egypt. Historically its flowers were used to prepare fabric dyes, food coloring, and for medicinal purposes. Most production today is for seeds used to extract its high quality edible oil, but there is also a market for seeds as bird feed (Li & Mündel, 1996). Although grown in numerous countries and environments worldwide, it remains a minor crop with average estimated production of about 800 000 t (Li & Mündel, 1996).

The USDA, Agricultural Research Service (ARS) maintains a collection of safflower germplasm at the Western Regional Plant Introduction Station (WRPIS), Pullman, WA, which currently includes about 2300 accessions. These accessions, representing germplasm from more than 50 countries, are available without charge to scientists worldwide upon request. Evaluation of these accessions for oil content and iodine number has been completed on about half the collection (Johnson et al., 1993). Additional information on oil content along with fatty acid composition

and other value-added factors such as vitamin E and phenolic glucosides is of interest for identifying accessions for value-added breeding and to assess the diversity of the USDA safflower collection.

In 1993 a safflower core collection representing about 10% of the total accessions was developed from the entire collection of more than 2 000 accessions based on country of origin and morphological data (Johnson et al., 1993). Because of limited evaluation data, oil and meal quality factors could not be considered in the development of the core collection. The objectives of the current work were (1) to complete an evaluation of 1 000 accessions from the USDA collection, including the core collection, for oil and meal factors, (2) to determine the extent to which the core had represented the diversity found in the 797 non-core accessions evaluated, and (3) to examine patterns of diversity for oil and meal factors among accessions from different geographic regions.

Materials and methods

In 1994, 1 000 safflower (*Carthamus tinctorius* L.) accessions from the USDA-ARS world collection were grown in field plots at the Montana State University-Eastern Agricultural Research Center, Sidney, MT, USA. This included the core collection of 203 accessions and 797 non-core accessions. The non-core accessions had limited oil evaluation information, and were those most recently added to the collection.

Each accession was planted in random order and grown in a single 6 m row, with rows 0.6 m apart. In order to maximize the total number of accessions evaluated and available for comparing core and non-core accessions, and for analysis among regions, individual accessions were not replicated. The commercial cultivars Montola 2001 (high oleic) and Morlin (high linoleic) were included as checks and planted alternatively every tenth row (54 total plots for each cultivar). A 1.8 m long section of each plot was covered with a mesh bag during anthesis to provide self-pollinated seed from about 20 plants. Seed from one selfed plant was analyzed for oil fatty acid quality. The remainder of the selfed seed was bulked and a representative sample of 16 g (approximately 500 seeds) of self-pollinated seed was used for analyses of oil content, tocopherols, and phenolic glucosides as described below.

Oil content. The 16 g sample of bulked safflower seed for each accession was dried at 60 °C for 4 h. A portion of this seed was ground and an accurately weighed sample extracted using a soxtec extraction system. The solvent used for the extraction was hexane and the extraction time was 1.5 h. The resulting oil extract was weighed and percent oil content determined. The extracted oil was frozen and stored in the dark for analysis of tocopherols, and the meal was dried and stored for analysis of phenolic glucosides as described below.

Tocopherol measurements. The vitamin E forms α- and β-tocopherols present in the safflower oil extract were measured for each accession using high performance liquid chromatography (HPLC) procedures developed by Waters Chromatography Division of Millipore Corporation (Milford, MA, USA). An aliquot of safflower oil was diluted one to ten with 0.3% isopropyl alcohol in isooctane and analyzed for the forms and quantity of tocopherols (mg per 100 g oil). An isocratic elution on a Waters (Milford, MA, USA) Nova-Pak Silica, 60 Å, 4 μm, 3.9 $\times$ 150 mm column at 35 °C and a flow rate of 1.0 ml/min was completed. With each chromatographic run of 40 samples, a set of concentration standards and an internal standard were included. The calibration samples of α-and β-tocopherols as well as the internal standard, 2,2,5,7,8-pentamethyl-6-hydroxychromin, were provided by Eisai USA Inc. (Teaneck, NJ, USA).

Phenolic glucoside measurements. The concentrations of the phenolic glucosides matairesinol monoglucoside (bitter) and 2-hydroxyarctiin (cathartic) in safflower meal derived from the 16 g samples from each accession used for oil extraction were measured using HPLC. It has been shown by Lyon et al. (1979) that the above glucosides are found only in safflower flour and not the hull. The defatted safflower meal was screened to remove safflower hull pieces from the seed flour and 100 mg flour was extracted with 3 ml of HPLC grade methanol containing kinetin-riboside as an internal standard. An aliquot of extract was diluted in half with water and analyzed for phenolic glucosides by HPLC.

A Waters (Milford, MA, USA) Nova-Pak C-18, 60 Å, 4 μm, 3.9 $\times$ 300 mm column maintained at 34 °C was used to affect separations. The solvent system included 0.02 M ammonium phosphate (solvent A, with phosphoric acid) and HPLC grade methanol (solvent B). A computer controlled gradient elution pattern started with a solvent A to B ratio of 69 to 31 for 28 min. The ratio was then adjusted to 10 to 90 over a 6 min period and held for 12 min before returning to the original ratio. Column eluate was monitored

for bitter and cathartic phenolic glucosides at 280 nm eluted over the first 30 min. Using authentic samples of matairesinol monoglucoside and 2-hydroxyarctiin, a set of concentration standards was developed and included with each chromatographic run. Peak areas for matairesinol monoglucoside and 2-hydroxyarctiin as compared with the internal standard were used to construct a concentration calibration to quantify the percent of phenolic glucosides in flour by weight.

Oil fatty acid determination. Three seeds from one of the safflower plants grown under the mesh bag were cut in half and placed in an aliquot of hexane. The cut seeds in hexane were crushed with a glass rod and an aliquot of hexane containing extracted oil was treated with sodium methoxide using procedures of Daun & Mazur (1983) and Liu (1994). The resulting methylated fatty acids were then analyzed using gas-liquid chromatography with a Hewlett-Packard 5890 gas chromatograph (Wilmington, DE, USA) and a J & W Scientific, DB 225, 15 m $\times$ 0.25 mm ID, 0.25 μm column (Folsom, CA, USA). Peak identification and response factors were determined using known samples of the appropriate methylated fatty acid esters, obtained from Nu-Chek Prep, Inc. (Elysian, MN, USA). The major fatty acids found were methyl esters of linoleic, oleic, palmitic, and stearic acid. The relative percentage of each ester was determined by integrating the area under each peak in the chromatogram. The area under each peak was found to be directly proportional to the percentage of the ester present, as confirmed by known standards.

Statistics. Accessions were divided into core and non-core groups. Means, maximum and minimum values, and variances of the groups were determined for each factor evaluated. The differences between core and non-core group means were compared using the F-ratio of the between group to the within group pooled variance with a significance level of $P<0.05$. Variances between core and non-core groups were compared using a modified Levene's test in which the absolute deviations from group medians were calculated and the resulting mean deviations compared using the F-ratio (Manly, 1986). The means and variances of the check cultivars (n=54) were also compared as above.

Accessions were organized into seven broad geographic regions as shown in Table 1. The regions were chosen to represent geographic areas without prior knowledge of the distribution of oil and meal characteristics. South-Central Asia, China, South-West Asia, the Mediterranean, and East Africa roughly corresponded to five of Vavilov's seven 'centers of origin' (Vavilov, 1997). Accessions from the Americas and Europe were introduced from other regions as evaluation and breeding programs were initiated in the first half of the 20th century (Knowles & Miller, 1965). A total of 29 miscellaneous accessions from various countries could not be assigned to a major region and were omitted from the analysis.

Regional means and variances were analyzed with general linear models as described above. Data were then standardized to have a mean of zero and unit variance and oil and meal data for regions analyzed using canonical discriminant analysis (CANDISC, SAS Institute, Cary, NC, USA).

Results and discussion

Core and non-core comparisons

The mean oil content for the 797 non-core accessions (PI's) in Table 2 was significantly higher ($P<0.05$) than the mean for the core collection PI's. This was attributed to the presence of numerous lines of improved germplasm in the non-core accessions acquired from the discontinued breeding programs of Dr. P.F. Knowles (University of California-Davis) and Dr. D. Rubis (University of Arizona) (Johnson et al., 1993). Mean values of linoleic acid, oleic acid, and β-tocopherol did not differ between the groups, and the variance differed only for palmitic acid. The core had higher mean palmitic acid, stearic acid, and cathartic phenolic glucosides than the non-core accessions, but lower α-tocopherols and bitter phenolic glucosides.

Means of linoleic and oleic fatty acids of Montola 2001 and Morlin differed as expected (Table 2), but the means for other fatty acids did not differ significantly between the cultivars. The variance for oleic and stearic acid was higher for Montola 2001 than Morlin, but this had no important effect on cultivar quality.

The range for oil content of the core collection and the non-core accessions was similar (Table 2). For linoleic acid, the maximum value was about 3% higher in the core than in the non-core accessions. The maximum oleic acid value for the core was about 5% less than the maximum for the non-core accessions. This was attributed to the presence of material in the non-core accessions selected by Knowles and Rubis for high oleic acid. The core had a maximum value for α-tocopherols of 119 compared to 160 mg/100 g oil for the non-core accessions (Table 2). For bitter phenolic glucosides the core had a substantially greater range

Table 1. Countries within regions used in analysis of safflower oil and meal factors

Americas	China	E. Africa	E. Europe	Mediterranean	S.C. Asia	S.W. Asia
Argentina (2)[a]	China (108)	Kenya (7)	Bulgaria (2)	Algeria (1)	Banglad. (12)	Afghanistan (8)
Canada (2)	N. Korea (1)	Eritrea (1)	Czech Rep. (1)	Cyprus (1)	India (363)	Armenia (2)
Mexico (4)		Ethiopia (4)	Hungary (3)	Egypt (27)	Pakistan (13)	Iran (96)
U.S.A. (160)		Sudan (16)	Poland (4)	Greece (1)		Iraq (3)
			Romania (1)	Israel (12)		Kazakhstan (6)
			Russia (1)[b]	Italy (3)		Kuwait (1)
			Ukraine (1)	Jordan (5)		Russia (1)[b]
				Libya (1)		Tajikistan (2)
				Morocco (5)		Uzbekistan (9)
				Portugal (8)		
				Spain (5)		
				Syria (7)		
				Turkey (61)		

[a] The number in parenthesis is the total accessions from each country within a region.
[b] Passport data indicated that one accession from Russia was geographically aligned with E. Europe and one with S.W. Asia.

than the non-core accessions, but the range of cathartic phenolic glucosides was considerably less for the core (5.52% in flour) than the non-core (10.01% in flour) accessions. So for % oil, linoleic, palmitic, and stearic acid, β-tocopherol, and bitter phenolic glucosides, the core appeared to capture as much, or more diversity than observed in the non-core accessions. But for α-tocopherol, cathartic phenolic glucosides, and to a lesser extent oleic acid, the core did not fully represent the range observed in the non-core accessions.

The maximum values for percent oil and linoleic acid for the core and non-core PI's were somewhat higher than that observed for Morlin, the high linoleic check (Table 2). And the maximum % oleic acid for Montola 2001 was higher than the highest value from the accessions. But for α-tocopherol and phenolic glucosides the range of values for the core and other accessions was considerably beyond the values for the cultivars (Table 2). It appeared, therefore, that the scope for substantial improvement in percent oil and either linoleic or oleic fatty acids using the germplasm collection was more limited than other value-added characteristics. This is reasonable because breeding programs have already concentrated on oil and fatty acid quality to a far greater extent than on tocopherols or phenolic glucosides.

Linear correlation coefficients were significant in two-thirds of the possible paired comparisons, and correlations among all the fatty acids were significant (Table 3). The negative correlation between % oil and linoleic acid may have been associated with the presence of improved lines with both high % oil and high oleic acid (thus low linoleic acid) donated by Knowles and Rubis. In general, there was a tendency for high % oil to be associated with lower linoleic, and higher oleic, palmitic, and stearic fatty acids. High % oil was also associated with lower α-tocopherol and higher cathartic phenolic glucosides (Table 3).

The strongest correlation was between linoleic and oleic acid (Table 3). Knowles (1969) described the genes involved in the inverse relationship between linoleic and oleic acid in safflower. In a study evaluating 200 safflower accessions, Fernández-Martínez et al. (1993) found that the strength and direction of correlations between certain fatty acids depended on the environment where the seed was grown. However, in their study, and in Table 3, correlations between oleic and linoleic acid content were always strongly negative, and the correlation between palmitic and stearic acid was always significantly positive. Thus those relationships were always consistent among different seed growth environments in their study and in this study.

Regional comparisons

Analysis of variance of regional means resulted in highly significant F ratios for each oil and meal factor in Table 4. However, the variance for each factor was also highly significant. Higher variance in a given region suggests more diversity for a given factor, but the differences in variance complicated comparisons among regional means because of heterogeneity of error (Steel & Torrie, 1980). Means for % oil were highest for accessions from the Americas, and this was

Table 2. Safflower seed analysis of 797 non-core plant introductions (PI's), the core collection of 203 PI's, and two check cultivars (n=54) from 1994 field plots at Sidney, MT, USA

	Mean	Minimum	Maximum	Variance[a]
Oil (%)				
797 PI's	29.1	13	46	20.93
Core PI's	26.8	14	44	18.96
Montola 2001	39.7	37	44	1.322
Morlin	40.8	37	43	1.264
Linoleic acid (%)				
797 PI's	76.8	11.0	83.1	103.4
Core PI's	77.3	12.1	86.5	87.6
Montola 2001	11.4	8.6	14.4	1.79
Morlin	80.8	78.0	82.6	1.19
Oleic acid (%)				
797 PI's	13.4	6.2	81.9	107.2
Core PI's	13.1	6.2	77.1	105.2
Montola 2001	81.1	76.9	84.4	2.71
Morlin	8.5	7.0	12.2	0.82
Palmitic acid (%)				
797 PI's	5.21	3.9	6.8	0.15
Core PI's	5.34	3.7	6.9	0.19
Montola 2001	4.12	3.8	5.3	0.21
Morlin	5.71	4.7	5.9	0.20
Stearic acid (%)				
797 PI's	2.09	1.1	4.5	0.11
Core	2.18	1.2	4.6	0.14
Montola 2001	1.03	0.8	1.9	0.24
Morlin	2.61	1.7	3.1	0.49
α-Tocopherol (mg/100 g oil)				
797 PI's	72.7	32	160	137.3
Core PI's	70.3	18	119	129.8
Montola 2001	63.9	51	91	46.0
Morlin	74.5	59	86	39.3
β-Tocopherol (mg/100 g oil)				
797 PI's	2.12	0	5	0.53
Core PI's	2.08	1	5	0.53
Montola 2001	1.78	1	4	0.30
Morlin	2.87	2	5	0.22
Phenolic glucosides, bitter (% in flour)				
797 PI's	0.48	0.17	1.30	0.025
Core PI's	0.38	0.02	2.29	0.033
Montola 2001	0.56	0.31	0.89	0.010
Morlin	0.57	0.33	0.84	0.012

Table 2. Continued

	Mean	Minimum	Maximum	Variance[a]
Phenolic glucosides, cathartic (% in flour)				
797 PI's	4.12	1.32	11.33	0.822
Core PI's	4.28	0.91	6.43	0.652
Montola 2001	5.45	3.26	7.31	0.515
Morlin	6.05	3.76	8.21	0.703

[a] The core and non-core variance is calculated from accessions within each group; the variance for the cultivars is calculated from the 54 replicate plots for each cultivar.

Table 3. Correlation matrix for nine oil and meal factors measured on 1 000 safflower accessions

	Oil[a]	Lino	Oleic	Palm	Stearic	α-Toco	β-Toco	Bitter	Cathartic
Oil	–								
Lino	-0.20^{**}	–							
Oleic	0.20^{**}	-0.98^{**}	–						
Palm	0.32^{**}	0.38^{**}	0.34^{**}	–					
Stearic	0.21^{**}	0.28^{**}	-0.30^{**}	0.34^{**}	–				
α-Toco	-0.16^{**}	-0.06	0.04	-0.18^{**}	-0.06	–			
β-Toco	-0.06	0.04	0.05	-0.01	-0.02	0.52^{**}	–		
Bitter	0.04	0.04	-0.04	-0.02	0.08^{*}	0.12^{**}	0.08^{**}	–	
Cathartic	0.31^{**}	0.10^{**}	-0.09^{*}	0.28^{**}	0.28^{**}	-0.21^{**}	-0.12^{**}	0.27^{**}	–

*,** Significant at P=0.05 and P=0.01 levels, respectively.
[a] Oil = % oil, lino = % linoleic fatty acid, oleic = % oleic fatty acid, palm = % palmitic fatty acid, stearic = % stearic fatty acid, α-toco = α-tocopherol (vitamin E, mg/100 g oil), β-toco=β-tocopherol (vitamin E, mg/100 g oil), bitter = % bitter phenolic glucosides (in flour), and cathartic = % cathartic phenolic glucosides (in flour).

again attributable to improved lines from the Knowles and Rubis breeding programs (Table 4). This also accounted for the low linoleic acid and high oleic acid means of accessions from the Americas. Because high oleic lines have low linoleic acid, they increased the regional mean for oleic and decreased it for linoleic acid in the Americas. Most of the high oleic acid germplasm was originally found in India and Bangladesh (Knowles & Mutwakil, 1963), accounting for the somewhat higher mean % oleic acid of accessions from South-Central Asia than from other regions, except the Americas. Because of the presence of both high oleic and linoleic lines, the variance for the Americas was especially high for linoleic and oleic acid. This was also seen to a lesser extent in South-Central Asia. High variance for α-tocopherols was found in accessions from China, the Mediterranean, and especially South-West Asia regions (Table 4).

The first and second canonical discriminant functions explained 80% of the total variation in oil and meal factors, 51% for function 1 and 29% for function 2. The analysis showed that accessions from the Americas, China, South-Central Asia, and South-West

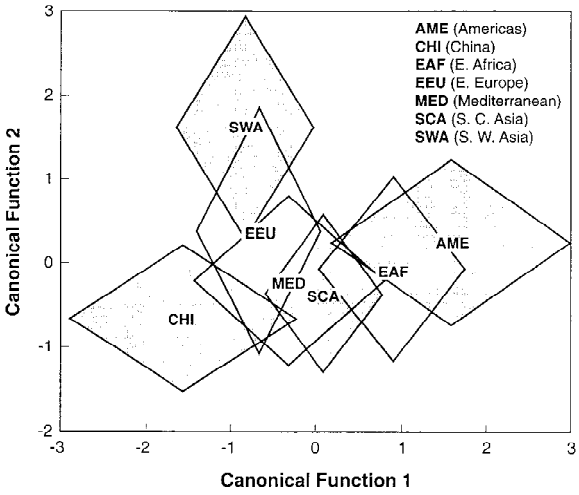

Figure 1. Plot of seven regions according to canonical discriminant functions obtained from analysis of nine oil and meal characteristics on 971 accessions. Diamond points indicate one standard deviation from the regional mean.

Asia could be differentiated fairly well based on oil and meal factors (Figure 1). But there was considerable overlap among accessions from East-Europe

Table 4. Means and variances for nine safflower oil and meal factors in seven geographic regions

		Oil[a]	Lino	Oleic	Palm	Stearic	α-Toco	β-Toco	Bitter	Cathartic
		—————%—————					— mg/100 g oil—		—% in flour—	
Americas	$\bar{x}$	33.6	73.3	17.3	5.46	2.19	71.1	2.01	0.43	4.03
n=168	s^2	29.7	314.2	350.4	0.21	0.17	87.3	0.47	0.01	0.40
China	$\bar{x}$	24.8	78.8	12.2	4.84	1.78	72.8	1.91	0.43	3.54
n=109	s^2	19.3	44.3	45.5	0.11	0.14	131.8	0.32	0.01	0.37
E. Africa	$\bar{x}$	30.5	78.9	11.2	5.61	2.23	65.6	2.00	0.35	4.70
n=28	s^2	7.4	0.6	0.5	0.17	0.05	29.8	0.37	0.01	0.26
E. Europe	$\bar{x}$	23.5	77.8	11.7	5.36	2.34	74.0	1.92	0.49	4.24
n=13	s^2	4.8	5.4	6.7	0.07	0.11	50.8	0.24	0.30	1.60
Med.	$\bar{x}$	27.0	79.3	10.6	5.28	2.16	70.7	1.99	0.40	4.34
n=137	s^2	12.5	3.3	1.9	0.17	0.08	150.9	0.35	0.02	0.72
S.C. Asia	$\bar{x}$	29.4	76.6	13.9	5.19	2.13	70.9	1.96	0.50	4.44
n=388	s^2	7.1	85.8	83.4	0.09	0.05	95.1	0.33	0.02	0.90
S.W. Asia	$\bar{x}$	25.8	77.8	11.4	5.28	2.01	79.8	2.91	0.52	3.65
n=128	s^2	7.5	38.2	42.9	0.10	0.05	265.8	0.84	0.04	0.58

[a] Oil = % oil, lino = % linoleic fatty acid, oleic = % oleic fatty acid, palm = % palmitic fatty acid, stearic = % stearic fatty acid, α-toco = α-tocopherol (vitamin E, mg/100 g oil), β-toco=β-tocopherol (vitamin E, mg/100 g oil), bitter = % bitter phenolic glucosides (in flour), and cathartic = % cathartic phenolic glucosides (in flour).

with those from South-West Asia. Accessions from the Mediterranean and South-Central Asia were closely aligned. The East African region, dominated by accessions from Sudan, was most closely aligned with the Americas. Many of the accessions first collected for testing in the USA were from the Sudan and two of these accessions became cultivars. They were N-9 (PI 542427), which was never established commercially, and N-852 (PI 572429), which was widely grown (Knowles, 1969). A single plant selected from N-852 (N-10, PI 572428) was widely grown (Williams, 1964) and used extensively as a parent of many US cultivars (Knowles, 1969).

For canonical function 1, standardized coefficients were highest for % oil and oleic acid, followed in importance by linoleic and palmitic acid. All coefficients for those factors were positive, so when those factors were high (Table 4), canonical function 1 also tended to be high (Figure 1). The high % oil, oleic, and palmitic acid for the accessions from the Americas contrasted with the relatively low values for these factors in China, leading to the wide separation between those regions. This separation is logical given the contrast in selection pressure between the traditional use of safflower in China — as a medicinal plant dating back over 2 000 years — and the major use of safflower in the Americas as an edible oil (Li & Mündel, 1996). The factors having the most influence on canonical function 2 were linoleic and oleic acids, which had negative coefficients, followed by β-tocopherol concentration and palmitic acid, which had positive coefficients. Thus the combination of average linoleic and palmitic acid values, relatively low oleic acid, but with very high β-tocopherol (Table 4) were important factors leading to the high canonical function 2 value observed for South-West Asia (Figure 1). In general, however, values for tocopherols and phenolic glucosides had relatively less influence on differentiating regions than did % oil and fatty acids.

The comparisons of the core and non-core accessions showed that although the core was not fully representative of the non-core material, it did capture a high fraction of the diversity present. Thus, the core offers a relatively small germplasm set useful for evaluation and screening of factors that may be too complex or expensive to complete on the whole collec-

tion. The canonical discriminate analysis showed that oil and meal factors among regions were sufficiently different to allow differentiation between regions in some cases, but not in others. Variation in % oil and fatty acids were generally more important than tocopherols and phenolic glucosides in differentiating accessions on a regional basis. Values for individual accessions for the oil and meal factors can be found using the Germplasm Resources Information Network (GRIN) (www.ars-grin.gov/npgs/). The GRIN system also includes data for other evaluation factors taken for safflower and can be used to order samples of specific accessions without charge.

Acknowledgments

The authors thank Chris Barrett for his technical assistance in data analysis.

References

Daun, J.K. & P.B. Mazur, 1983. Use of gas liquid chromatography for monitoring the fatty acid composition of Canadian rapeseed. J. Am. Oil Chem. Soc. 60: 1751–1754.

Fernández-Martínez, J., M. del Rio & A. de Haro, 1993. Survey of safflower *(Carthamus tinctorius* L.) germplasm for variants in fatty acid composition and other seed characters. Euphytica 69: 115–122.

Johnson, R.C., D.M. Stout & V.L Bradley, 1993. The U.S. Collection: A rich source of safflower germplasm, pp. 202–208. In: Li Dajue and Han Yuanzhou (Eds.), Proceedings Third International Safflower Conference — 18 June 1993, Beijing Botanical Garden, Institute of Botany, Chinese Academy of Sciences, Beijing, China.

Knowles, P.F., 1969. Modification of quantity and quality of safflower oil through plant breeding. J. Am. Oil Chem. Soc. 46: 130–132.

Knowles, P.F. & M.D. Miller, 1965. Safflower. California Agricultural Experiment Station Extension Service Circular 532. University of California.

Knowles, P.F. & A. Mutwakil, 1963. Inheritance of low iodine value of safflower from India. Economic Bot. 17: 139–145.

Li, Dajue & H.-H. Mündel, 1996. Safflower. *Carthamus tinctorius* L. Promoting the conservation and use of underutilized and neglected crops. 7. Institute of Plant Genetics and Crop Plant Research, Gatersleben, Germany/International Plant Genetic Resources Institute, Rome, Italy.

Liu, K., 1994. Preparation of fatty acid methyl esters for gas-chromatographic analysis of lipids in biological materials. J. Am. Oil Chem. Soc. 71: 1179.

Lyon, C.K., M.R. Gumbmann, A.A Betschart, D.J. Robbins & R.M. Saunders, 1979. Removal of deleterious glucosides from safflower meal. J. Am. Oil Chem. Soc. 56: 560–564.

Manly, B.F.J., 1986. Multivariate statistical methods A PRIMER. First ed., Chapman & Hall, London.

SAS Institute. 1985. User's Guide: Statistics. 5th ed. SAS Institute, Cary, NC, 1985.

Steel, R.G.D. & J.H. Torrie, 1980. Principals and procedures of statistics. Second ed., McGraw-Hill, New York, NY.

Weiss, E.A., 1971. Castor sesame, and safflower. Barnes and Noble, Inc., New York, NY.

Williams, J.H., 1964. Registration of N-10 Safflower. Crop Sci. 4: 446.

Vavilov, N.I., 1997. Five Continents. International Plant Genetic Resources Institute, Rome, Italy.